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(54) Title: ELECTRODE STRIPS FOR TESTING SMALL VOLUMES

(57) Abstract

A test strip comprising a support carries an active electrode and a counterelectrode, and a layer of material within which a small volume of liquid to be tested can be distributed and provide contact between the electrodes, and wherein an analyte-specific reagent is coated on the material. The layer of material can conveniently be provided in the form of a tape from which sections can be cut or used sequentially.

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Electrode Strips for Testing Small Volumes

Field of the Invention

This invention relates to electrode strips for testing small volumes of, say, whole blood.

Background of the Invention

Diabetes is one of the most common endocrine conditions. Sufferers must monitor their blood glucose level frequently. This is usually achieved by the use of small test strips which detect blood glucose.

Problems commonly experienced by users of these test strips are an inadequate amount of blood on the test strip and bad placement of the blood on the test strip. A number of devices have addressed this problem by using sample chambers that fill by capillary action. The sample is retained in close proximity to the electrodes which facilitate the measurement of the specific analyte in the sample; see EP-A-0170375 and US-A-5141868.

Such known devices comprise electrodes deposited on a non-conducting substrate, coated with a reagent system specific for the analyte of interest and housed within a cavity whose dimensions are sufficiently small to allow introduction of a sample, e.g. 2.5-3 µL in volume, by capillary action. The extent to which these devices can be miniaturized is limited by both the manufacturing tolerances and the signal-to-noise ratio achievable with their chemistry.

US-5820551 discloses a test strip comprising a support carrying a working electrode and a counter electrode, and an enzyme and a mediator that are coated on the active electrode. A drop of whole blood can provide a conducting path between the electrodes, and the concentration of glucose in the blood can be determined. The active electrode is exposed to a whole blood sample without an intervening membrane or other whole blood filter.

WO-A-98/55856 (published after the priority date claimed for this Application) discloses an analyte-specific reagent coated on the conductive layer, and a monofilament mesh laid over the reagent and the reference electrode. A sample application area is provided at one edge of the mesh.

Summary of the Invention

According to a first aspect of the present invention, a test strip comprises a support carrying an active electrode and a counterelectrode, and a layer of a material within which a small volume of liquid to be tested can be distributed and provide contact between the electrodes, and wherein an analyte-specific reagent such as one component of a redox reaction, e.g. an enzyme, co-factor or mediator, is coated on the material. In particular, the invention provides a test strip for blood glucose, in which the sample requirement is very small, and efficient reaction kinetics are achieved by the application of the reagents in a novel manner.

The reagent-coated material may itself be in tape form. According to a second aspect of the invention, a flexible tape is of a material within which liquid can be distributed and on which are coated discrete areas of at least one component of a redox reaction.

Description of the Invention

In accordance with this invention, any one or more of the components of a redox reaction, e.g. an enzyme such as glucose oxidase or glucose dehydrogenase, a co-factor and a mediator may be applied to a mesh or membrane which is placed over the device. For the purpose of illustration only, the invention may be described with reference to an enzyme-coated mesh. Whichever component or components are used, when the sample is added, they are solubilised quickly and form an efficient reaction medium that can provide contact between the separate electrodes of the test strip. In this manner, the reaction will proceed rapidly and without diffusion barriers. This reaction configuration is particularly indicated in cases where the sample volume is low, the sample is viscous (such as with whole blood) and a rapid reaction is required.

In a typical embodiment of the invention, the sensor test strip consists of two electrodes, one of which acts as a working electrode and another which acts as a counter, reference electrode. The end of the working electrode that is exposed to the sample has a mediator in intimate contact with it. The test strip effectively provides a reaction chamber defined by these two electrodes

and an additional sheet, overlying the electrodes, which has been pre-coated with the redox enzyme and any necessary co-factor for that enzyme. The reaction chamber may also comprise further sheets of material and/or wetting agents, e.g. a surfactant, or cell-lysing materials (which may be placed in any one of the overlying sheets). In this manner, the active enzyme is not coated onto the conductor which forms the working electrode but is provided in a separate layer above it which, in turn, effectively forms the solution phase of the reaction chamber. When combined with lateral flow, conditions are created that approach efficient mixing in a stirred reaction chamber.

In an example of the invention, a silver chloride/silver reference/counter electrode is located adjacent to a carbon electrode. Typically, for this purpose, a pair of printed carbon electrodes is printed on a non-conducting substrate, and then silver/silver chloride is printed on one of the carbon electrodes to function as the reference/counter electrode. A non-conducting ink is printed over the carbon electrodes and the substrate, in order to define a portion of each electrode as a contact pad for insertion into a meter and another portion on each electrode away from the contact pad as the sensing area where the sample is received.

A mediator for the enzyme cofactor NADH is then prepared and deposited onto the electrode from aqueous solution by pipetting. A further layer containing NAD is then deposited onto the working electrode.

A monofilament mesh material is coated with a surfactant and then with a solution containing glucose dehydrogenase via pipetting, ink jet-coating or dip-coating, and is placed over the two electrodes to form a reaction chamber. This reaction chamber may be defined further by additional printing, or by the use of a top layer to form an edge fill cavity. For example, a second non-conducting ink printed on top of the mesh material, and then a cover tape is applied on top of the mesh in such a way as to leave an extended area of the mesh exposed for sample application.

The device allows the application of a small volume of sample (typically 1 µL or less) to the mesh extension. This is followed by flooding of the device

sensing area with sample, bringing it into intimate contact with the measuring electrodes.

Devices having an edge fill are described in WO-A-98/55856. They can be simply adapted, in accordance with the present invention. In particular, reference may be made to Fig. 1 in WO-A-98/55856; components of this invention are the support (1), electrodes (2/3), mesh material (6) and tape (7); in addition, reagent is provided on the mesh material. Such a device can work by application at its edge, to a sample. This is particularly valuable in cases where it is difficult to extract the sample. Other configurations will be evident to one skilled in the art, including combinations of one or more of the cofactor, mediator or the enzyme coated onto the overlying mesh or membrane sheets. The choice of combination may on the reaction kinetics of the various compounds.

In another embodiment of the device, the enzyme or the mediator is coated on the sheet, the co-factor and the other of the mediator or the enzyme are coated onto the working electrode directly, and the sheet is capable of filtering the whole blood such that the active electrode sees a sample which is effectively free of whole blood cells. In this case, the haematocrit dependency of the result is substantially reduced. In this manner, the cell-filtering function of a selected membrane may be combined with the rapid kinetics of having the some or all of the active elements of the reaction (the enzyme, mediator and the co-factor) in the membrane, to produce a highly effective device.

In summary, according to the present invention, a device is constructed by depositing one or more of the reagents required for the quantitation of an analyte as a single or multiple layers on a fine mesh material or membrane; the deposited areas are of dimensions small enough to wet with a very small sample volume. The mesh or membrane can be used in both colorimetric and electrochemical devices.

A characteristic of this invention is that a reagent is applied precisely onto a target area on a woven material such as polyester or nylon or other porous membrane. In use, this provides rapid solubilisation of the reagents in the presence of the sample. The reagent or reagents can be applied in a

number of different methods that result in the deposition of a known volume at a precise location and in a well-defined foot-print. These include the use of dispensing equipment such as a piston pump, syringe pump or on-demand ink-jet printer.

In a further embodiment, a flexible tape containing one or more reagents may be laminated to another flexible tape on which is printed a series of electrodes. Instead of cutting out individual sensors, the laminate (comprising a row or series of sensors) may be used sequentially, e.g. on being dispensed from a suitable dispenser. For this purpose, whether or not as a laminate, a tape of the invention may be provided as a roll, and stored in sealed cassettes which may also contain desiccant. In use, the cassette may be inserted into a automatic dispenser from which the tape is wound out automatically by an indexing mechanism to reveal sequentially the discrete sensors. The action of this instrument is therefore analogous to the action of a film in a camera. In this embodiment, the tape may also contain a red blood cell-lysing reagent such as saponin, in order to reduce the effect of haematocrit and haemoglobin in a whole blood sample. The tape may be further protected from moisture by being covered with a peelable film (e.g. of aluminium) that is automatically peeled off when the tape is dispensed from the cassette. When the sample is applied to the sensor, the amount of analyte of interest in the sample may be determined electrochemically. Such determination can be conducted by known methods.

The following Example illustrates the invention.

Example

A conductive ink material is printed onto a non-conducting polyester sheet material by a screen-printing process. The conductive ink material consists of a mixture of graphite and carbon particles and a polymer binder in an organic solvent. After deposition of the conductive ink, solvents are removed in a forced air oven. A silver/silver chloride reference/counter electrode is printed onto one of each pair of printed carbon electrodes followed by a non-conducting ink layer to define the contact pads and the sensor area.

A mediator such as Meldola Blue, Nile Blue or other suitable dye and the enzyme co-factor nicotinamide adenine dinucleotide (NAD) are deposited onto

the carbon electrode. Alternatively, the NAD is applied separately over the mediator from an aqueous ink.

The enzyme glucose dehydrogenase is deposited as uniform spots on a monofilament polyester mesh tape. This is achieved as follows:

(a) in a contact mode, where a drop formed at a dispenser tip in close proximity to the mesh is allowed to be transferred to the mesh by touching off the drop onto the mesh surface; or

(b) in a non-contact mode, where a drop formed by an ink-jet print-head or other orifice above the mesh is dropped onto the mesh from a distance under conditions which do not cause it to penetrate the mesh.

Upon drying, the spots spread to cover an area defined partly by the characteristics of the mesh weave and partly by the application conditions. Typically the areas covered by a 500 nL drop is 1.3 x 1.2 mm. The mesh tape is allowed to dry at room temperature.

The enzyme-modified mesh tape is then laminated onto the modified sheet of devices and secured further by a non-conducting print. Finally, a cover tape is laminated on tope of the mesh. The sheets of devices are disc cut into individual devices. In an alternative device format, the laminated sheets are wound and included in a cassette type unit, allowing a single device to be used by a wind-on mechanism similar to a camera film-winding system.

CLAIMS

1. A test strip comprising a support carrying an active electrode and a counterelectrode, and a layer of material within which a small volume of liquid to be tested can be distributed and provide contact between the electrodes, and wherein an analyte-specific reagent is coated on the material.

2. A test strip according to claim 1, wherein the reagent is at least one component of a redox reaction, e.g. one or more of an enzyme, a mediator and/or co-factor for the enzyme.

3. A test strip according to claim 2, wherein the at least one component comprises the enzyme.

4. A test strip according to claim 2 or claim 3, wherein the enzyme is glucose oxidase or glucose dehydrogenase.

5. A test strip according to any preceding claim, wherein the material is a monofilament mesh or membrane.

6. A flexible tape of a material within which liquid can be distributed and on which are coated discrete areas of at least one component of a redox reaction.

7. A flexible tape according to claim 6, wherein the material is a monofilament mesh or membrane.

8. A container containing a wound tape according to claim 6 or claim 7.

9. A container according to claim 8 also comprising automatic dispensing means.

10. A method for testing a liquid for the presence of an analyte, which comprises contacting the liquid with a test strip according to any of claims 1 to 5, and detecting the current.

11. A method according to claim 10, wherein the liquid is blood and the analyte is glucose.

INTERNATIONAL SEARCH REPORT

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A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C12Q1/00 G01N35/00 G01N27/30 G01N27/327

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C12Q G01N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

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[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

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